Some observations on Water Analysis - Chemical and Bacteriological.

Chemical Analysis, although popularly believed, and particularly so by Local Authorities, to be able to detect pollution and give a verdict for or against the use of a water for domestic purposes, has long since been denied such powers by trained Sanitarians. It has, however, maintained its place in determining the quality of a water, and a Local Authority and its Medical Officer of Health still look upon the chemical Report of a water Analysis as a most important factor. The bacter-iologist has not managed to come up to expectations yet, and even when disease has been sown broad-cast in many epidemics by means of water, he has been unable to isolate the organism and the failure has been to the detriment of the new Science. In fact many are of the belief that a minute supervisory control and inspection of the sources &c of a water supply would yield more certain results so far as pollution, than either a Chemical or Bacteriological Examination. The minds therefore of those who have to guide Local Authorities are much exercised over the question. They yearn for something definite to assist them in coming to a decision. They feel that the main responsibility lies on them, that they ought to submit an opinion based, not on a Chemical Analysis alone, but on a much broader survey of the question. The object/

object of a water analysis is to gain information regarding the quality of a certain water. The quality is not a constant, and varies according to many well-known conditions. But unless the Analyses are for the purpose of selecting, or choosing between different schemes for a new Water Supply, the ordinary information desired is whether pollution is obtaining entrance into a Water Supply, and more especially pollution of animal origin. This is very noticable in the presence of an epidemic believed to have a water origin. Till within a few years ago Chemical Analysis was depended upon solely for a verdict on this point, and the opinion of the Analyst was based on the amounts of certain ingredients contained in the sample. Much difference of opinion existed among Chemists not only in the methods employed but of the degree of significance that ought to be attached to the results obtained. Up to the present moment such conditions still obtain as a reference to latest volumes of the Journal of the Society of Chemical Industries, will show. It is many years ago since Dr. Buchanan wrote "We must go beyond the laboratory (meaning Chemical laboratory) for evidence of any drinking water being free from dangerous organic pollutions", and that "Chemists can tell us of impurity and hazard, but not of purity and safety". I regret to say that from personal experience some Chemists either forget, ignore, or are ignorant of such/

such words of wisdom, and consequently issue reports and opinions, the latter of which ~~is~~ are no logical sequence of the former. No fault can be taken with the care Analysis is carried out, or the ability of Chemists to discover minimal quantities of ingredients. Wankyln's Test is said to discover 1 part in 10,000,000 times its weight of water (Wanklyn and Chapman's Water Analysis). Erdmann's Test for Nitrates-which Wankyln always maintained was of much importance along with his process-can readily detect 1 part in 200,000,000, and having an upward limits of probably 1000,000,000. (H.Causse Compt~~is~~es Rendus 130 p. 785), Recognising these minute chemical tests, biological methods of water examination are now appealed to by many as a much more delicate and certain method for detecting pollution. It therefore seemed to me as a suitable- and probably profitable so far as acquiring definite information- subject to investigate, and if possible determine what was the present day position of methods of Water Examination for detection of pollution. The result of these investigations, together with the generalised notions resulting from the practical examination of nearly 250 samples of water, I now present as a thesis for my M.D.degree.

I trust that no apology is necessary for the statement that a supply of pure water is of supreme importance to any community. It is one of the most powerful public health defences we possess. Facts and statistics/

statistics are multitudinous in Sanitary literature demonstrating the beneficial effects of an abundant supply of wholesome water. The necessary sewerage system which requires to be provided, for a considerable time got much credit as being perhaps the principal factor in the diminution of mortality, but already the pendulum has swung back far enough to recognise the true cause - an abundant wholesome water supply. In Scotland it is a duty imposed upon a Local Authority to see that every occupied house in its district is provided with a proper supply of wholesome water at or reasonably near it.(Section 125 Public Health (Scotland) Act, 1897.) My paper is not an exhaustive one into various means by which contamination of a drinking water may occur. I exclude all other process but pollution by sewage, but in passing I may point out the present position of plumbo-solvent waters, and to remark that some allowance should be made to those workers in the domain of organic compounds, when the etiology of an inorganic pollution is not definitely settled.

My stand-point enables me to dispense with any consideration of Total Solids, Hardness, and Chlorides. The latter item on which much stress was put in the past, is now known to be of subsidiary importance, as a guide to pollution. In my own district there are two springs within ¾ of a mile of each other, and/

and both distant miles from a human habitation, or cultivated fields, one of which yield ·7 parts per 100,000, and the other 5·8 parts per 100,000. The chlorine factor in chemical analysis can only be of value when the chlorine factor of the adjacent streams is known, and even then as in my illustration without examination of the topographical and geological conditions obtaining in the part, a wrong conclusion may be arrived at. The chlorine item in a water analysis is only of value as an index showing urine pollution, a fact not always kept in mind by laboratory chemists.

Chemical Analysis has undoubtedly existed for a very long time, but in the 1st edition of Wankyln & Chapman's book on "Water Analysis" published 1868 they declare it to be the first book published on that subject. His famous process for estimating organic matters in water is given in that book. Previously such matters had been estimated by the difference of weight caused by the ignition of the total solids, and the colour of the residue also was regarded as of importance. In one of Wankyln's later editions he remarks that such a process "is now regarded as a curious eccentricity".

It says a good deal for Wankyln and his process that 34 years after its presentation to the chemical world, it has not undergone any improvement, and remains still the most practicable and widely used process in analytical laboratories for determining the/

the amount of organic matter in waters. Unfortunately this is not due to its absolute accuracy in revealing the amount of organic matters present. Wankyln laid down, in the 3rd edition of his work, standards for waters ranging from "High organic purity" to "Polluted", and by these standards the great majority of analytical chemists have formed judgments regarding a water under consideration. Free Ammonia present in very small amount and Albumenoid Ammonia ·010 per 100,000 condemns a water for domestic purposes by Wankyln's standard. Modern chemists have spoken strongly against the adoption of any general standards of purity, as laid down by some chemists, declaring it to be absolutely dangerous. I may instance Adams, Hehner, Dupre, Thresh &c. They believe that waters ought to be judged by local standards of purity , that is of unpolluted waters of the district from which the sample has come. Wankyln says "The analytical characters as brought out by the Ammonia process are very distinctive of <u>good</u> and <u>bad</u> waters, and are quite unmistakable". It shows that he himself clearly apprehended that his process was effective only at the extreme ends, and that for a pure water fairly polluted his process was not sufficient for detection. Drs. Cory and Dupre carried out many investigations for the Medical Department of the Local Government Board (11th Annual Report 1881-82.), with special reference to experimental/

tal contamination of waters. They found that extremely different quantities of nitrogenous polluting materials, may furnish equal amounts of Albumenoid Ammonia. The converse was also true. He shows that 1 gr. of human excrement per gallon of distilled water yielded ·014 parts per 100,000 of Albumenoid Ammonia. A result also obtained was that a healthy stool gave more Albumenoid Ammonia than one from a diseased person, the conditions as to food, &c. in the two cases being alike. In another experiment an amount of specially polluting matter was introduced into a quantity of water far in excess of what had obtained in the Caterham Epidemic, but beyond slightly increasing the Albumenoid Ammonia no indication was given of its highly objectionable and dangerous character. From such experiments it was plainly seen that Chemistry must progress, and if possible in the direction of exposing or separating organic matters of a harmful character from that of a harmless. Twenty years have elapsed since these researches and chemistry has not made any notable advance. A number of epidemics of Typhoid Fever in England which have been believed by the Medical Inspectors of the Local Government Board to be due to specially polluted waters are given in Dr. Thresh's book on Water Supplies p.177. Such remarks as a "further illustration of the inability of a chemist to prove the quality of organic matter in water when its quantity is small". (Buckingham)/

Buck ingham Outbreak 1888), again Dr Thorne on the investigations of Dr Barry declared that "Seldom if ever has the proof of the relation of the use of the water so befouled to wholesale occurence of Typhoid Fever been more obvious or patent". (Tees Valley Epidemic 1890). In connection with water from Dr. Frankland downward many chemists certified it as free from any trace of sewage. Such was also certified of Beverley Outbreak 1884, of the Trent 1893 and Worthing in 1893.

Thresh gives as his opinion (Wallis Ruppolico) (1895) that "by no process can either the quality or the quantity of the organic matter in water be determined". This is due to complexity of organic compounds and although even the quantity was able to be accurately determined, it would only afford the knowledge that so much pabulum for organisms was dissolved in the water, but of its source or origin-the principal question-we would be little better than we are now. I am not contending for chemistry to be able to detect typhoid excreta let us say in water, but chemical analysis must remain unsatisfactory, deficient, till it is able to detect sewage. I am aware that attempts to directly trace different ingredients that are known to be excreted by man in faeces and urine have been made, but in no casehas it been found practicable to utilise them so far as water pollution is concerned. Skatol, Indol &c have thus been investigated. The most recent is to/

to trace Cystin as a test of comtaminated water of animal origin. It certainly gives one a fair idea of much of the groping in research work, and is to my mind a painful example of what one has to peruse as a result of a fanciful title, in an earnest hope that the goal has at last been reached. This article is in the Bul.de la Soc. Chimique de Paris 1900 p.481 entitled "Sur la presence de la Cystine et de la Tyrosine dans les eaux contaminees". A method is then described for the direct examination of water for sewage contamination. As a student of practical physiology, I well remember the difficulty we experienced in obtaining Cystin and Tyrosin from much more comcentrated solutions than a contaminated water supply would ever possibly be. But Cystin and Tyrosin are generally pathological products, or at least products of retrograde metamorphosis. To get the large quantity necessary the author boldly affirms that Cystin is a product of decompositions of proteid matter in sewage. The test is a colorometric one, the test solution chloro-mercurate of amino-suphonate of soda. The reaction is due to the formation of a cystinate of iron. Foularton and Kellas in Vol. Xll of Public Health describe experiments as having been instituted by them to verify Causse's experiments. This they were unable to do, and further remarked that Causse's test solution was not even a test for Cystin. It is true

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that Cystinate of iron has not been isolated from the samples by Causse. It scarcely requires to be stated that Foularton and Kellas regard this test as valueless for practical water analysis. So much for direct Chemical Analysis of polluted waters.

Reverting again to Wankyln's Ammonia process, the amount of Nitrogen which leaves the organic matter (and of which in animal matters it forms 16% although in vegetable matters much less) to form the Albumenoid Ammonia varies as to the amount so let loose. The Massachusetts Board of Health believed that one-half of the nitrogen is converted to Ammonia as they carry out the process, but all admit its inconstancy except in the same sample. Probably only that portion which has progressed so far in the process of physico-chemical change of organic matter is so acted on to give off its nitrogen atoms, and this may explain the greater readiness of animal matters to give off Albumenoid Ammonia than vegetable, the former being so much more prone to change. One point agreed by all is that the amount of Albumenoid Ammonia is not an index to the amount of organic matter in the water.

Comparison of the same sample with other processes having in view the same object has not been regarded as satisfactory. This is, so far as determining the total amount of organic matter, a factor which even if obtained is not to be compared in a hygienic light, with the value which would be derived from a knowledge of/

of its animal or vegetable origin.

Frankland's ingenious method, which is so complex and subject to such corrections, that I should think that only a very few besides Frankland would risk their final figures as being accurate, may be put out of calculation although the results were not so far as published such as to bear any constant ratio. The latest attempt to ratio the Albumenoid Ammonia and Kjeldahl process (Jour Soc.Chem.Indus Vol.16 p.995) is clearly unsuccessful as the widely discordant figures show:-

Alb.Amm.	Kjeldahl.
·120	·50
·267	·85
·073	·31

Tidy's process also gives only comparative results and does not in any way indicate the amount of organic matter present. Like the Ammonia process the results are only of value when the organic matter in the various samples has the same composition. Its own composition determines the readiness of the oxidation. Wolff-Degener-Herzfeed discovered the complicated and long process of oxidising Potassium Bichromate, which yields a higher per centage of organic matter than the Tidy. This may be a step nearer the estimation of the total amount but even if that was gained ~~the~~ its significance is more apparent than real.. In fact out of the difference between the results of the two processes/

processes Barnes in the Journal of the American Chemical Society (1898 Ithink) proposes an improved method. The ratio, he points out, is generally less than unity. Woodman in the same Journal repeated these experiments and verified them. He states that "the ratio is constant if the character of the organic matters does not change and is independent of the amount present". In peaty waters the ratio is higher than .6 in sewage and sewage comtaminated waters ~~higher~~ lower than ·6.

Dr. Houston in his experiments for the Local Government Board on "soil washings" contented himself with Chemical Analyses showing the Free and Albumenoid Ammonias and the oxygen absorbed as being sufficient points upon which to obtain knowledge of purity or pollution. He showed that "soil washings" do not very greatly alter the composition of the liquid in respect of organic matter. His results even showed that from virgin soils much higher results were obtained than from polluted soils rich in animal organic matter. Now it is generally conceded that the dangerous pollution is organic in its composition, that from sewage the most, from peat the least. Even as regards the latter Wankyln says in the last edition of "Water Analysis" 1896 p. 71, "Albumenoid Ammonia over 1.5 per 1,000,000 ought to condemn a water absolutely.....it would be a great mistake to allow water highly contaminated with vegetable matter to be taken for domestic/

domestic use."

While admitting the difficulty of mathematically proving such a statement, I have sought in vain for any logical reasoning on this subject in any of the standard works and published papers of those who have written on this matter. Positive statements which ought to be only founded on such reasoning are abundant. Wanklyn's amount of organic matter in water to absolutely condemn, quite irrespective of its source, is just the amount that Dr Cory showed to be contained in one gr.of excrement in a gallon of distilled water. This is a concrete fact to grasp, and even granting that two grains of vegetable matter were equal to one of animal in its amount of nitrogen in its organic matter, an upland surface water containing this amount is to be condemned. The slightest amount of organic matters of animal origin in a drinking water ought to condemn it, but the process of chemical analysis is unable to capture red-handed such an intruder, and then commences to over-step its limits both experimental and empirical. That such a small amount as two grains of dissolved vegetable matters per gallon would induce an unhealthy condition in a human being who drinks on an average ·33 gals (Parkes) per day is scarcely in accordance with commonsense. A paper was read at Aberdeen Public Health Congress in 1900 by the City Analyst for Aberdeen (Journal State Medicine 1901 p.185) on "the effect on health of

organic/

organic matter in water" and as a specimen of unconsequential reasoning it would be hard to beat. As he has no claim to be heard on questions of disease in human beings, I do not trouble with any of his views, but would point out in passing that on the subject of his paper he does not adduce a single logical evidence in support of his contention. No more trustworthy or experimental basis for the conclusion adduced is to be found than when such statements were first hazarded, and the casual relationship between diarrhoea and organic matter in a water which would chemically be condemned is not only not proved, but is more probable to be erroneous. Even the idea of the more readily oxidisable matters being probably the most dangerous is only a pious opinion. In the 1860 Report of the Medical Officer of the Privy Council p.75 is described a well analysis in Coventry where the diarrhoea is ascribed to the organic matter, which according to the process of estimation at that time showed 8·1 parts per 100,000. But the total solids amount to 158 parts per 100,000, and the permanent hardness to 74° (metrical scale). To say that the organic matter was the producer of disease or illness in this case is unwarranted, although the chemical analysis clearly shows an unsuitable drinking water supply. In this instance I am merely decrying the want of scientific method in assigning to an impurity a cause of illness which might be shared if not principally caused by other ingredients. All know the story of the "Argo" with/

with its stricken passengers and untouched crew, and it has served for two generations to incriminate marshy water as a cause of malaria, and then as afterwards - and as it does in connection with typhoid at the present day - the unhesitating positive statement acts as a balm to troubled minds, and satisfies and lulls to rest. To prove the entrance of pollution to a water supply, the consumers of which are being visited by an outbreak of typhoid, is sufficient in all reports I have seen to declare it as the likely cause, which at the next step becomes the cause. The significance of such a discovery as an etiological factor of an outbreak is that all minds are instantly eased. Sometimes a water supply and its distribution is sanitarily perfect. As there are now many water supplies that are under excellent supervisory control and the origin of typhoid in such are more indeterminate. It will in my opinion be from the latter class that the truest advance in the etiology and origin of Typhoid Fever will be derived.

Regarding these inorganic salts which are believed to be an oxidation of nitrogenous organic matter - the Nitrates and Nitrites - I would state that so far as appears from papers in the chemical press, these are the substances that chemists are anxious to discover the process for their minute estimation. Nitrates were looked upon by Wanklyn as of considerable value in judging of the quality of a water. Still our knowledge of the nitrifying or denitrifying/

denitrifying action of micro-organism is far from perfect. Professor Warington maintains that the formation of nitrates in the soil is due to nitric organism which the soil always contains and they are always present in river water and sewage. These nitro-bacteria include nitrous and nitric organisms which seem to be identical in almost every respect except their chemical action or function. Muntz who took up the hint thrown out by Pasteur that the production of nitrates might be due to germ activity, still maintains that their action is limited to the oxidation of nitrogenous matter to nitrites, the further conversion to nitrates being due to the joint action of carbonic acid and oxygen. However so far as the relation of Nitrates and Nitrites to water analysis is concerned the fact remains that organisms capable of producing these salts are widely disturbed in nature, and in addition there are other ways of production of these nitrates and nitrites than by bacterial action. Schonbein says "the presence or absence of nitrites must not be regarded as the conclusive criterion of the purity of the water." Causse (Comptes.Rend. 130 p.785) says that good water should contain no nitrites, and that they are present in all animal contaminated waters, being produced by pathogenic bacteria. Beijerinck (Centrall für Bakt: Vol 12 p.715) states that cholera cultures which give the indol reaction on the addition of acid often do not contain nitrites. He examined in 1885 the Spree waters/

waters and isolated organisms from it which produced nitrites, but the water did not give a reaction to the applied tests for nitrites. Nitrites of microbial origin are produced far more from the vital activity of saprophytic forms than pathogenic germs. If water is kept in a bottle for sometime the ammonias diminish more especially the free ammonia, and the nitrates and nitrites proportionally increase. There is always the same amount of nitrogen compounds, only redistributed. It matters not under what conditions the water is kept the same results will follow. It may be kept at rest, or frequently agitated, in the dark or exposed to light, the bottle may be partially or completely filled. Frankland has shown that the micro-organisms increase enormously for a time under like conditions. In the Rivers Pollution Commissioners Report it is stated that upland surface waters in contact only with mineral matters or with the vegetable matter of uncultivated soil contain if any mere traces of nitrates and nitrites, but as soon as the water comes in contact with cultivated land, or is polluted by the drainage from farmyards or human habitations nitrates in abundance make their appearance. It has long been known that certain deep chalk wells contain much nitrates, but no sufficient explanation has yet been given to account for their presence. As much as one grain of nitric nitrogen per gallon has been found by Thresh in Norfolk in

waters absolutely beyond possibility of contamination by recent sewage. Nitrates being probably the final oxidation limit, it shows that self purification has resulted, and can give therefore no indication of the time of pollution. In Essex many deep chalk wells do not yield any trace of nitrates. But authorities have laid down standards by which to pass or condemn a water so far as nitrates are concerned.

To offer an opinion on a solitary water sample as is so frequently done, must be a hazardous proceeding. Much harm has undoubtedly been done by chemical analysts by their dogmatic assertions regarding the suitability or unsuitability of a water for domestic purposes. A single Analysis (except in the case of a deep spring) may be very misleading, but to carry such proceeding over a sufficient space of time is generally impracticable. I have therefore come to the conclusion that an analyst should not be required to give an opinion regarding a water submitted to him say by a Local Authority. All that can conscientiously be expected of him is, to estimate the quantities as far as possible or usual of the various ingredients, and with that completed his work ought also to end. F.Abba in Proc.Instit.Civil Engineers, 1900, makes the following statement "No investigation of water of unknown origin should be undertaken nor of samples procured by unreliable observers". The chemist/

chemist must thus be dependent on second-hand evidence for part of his opinion, and to my mind the estimate of the quantities of ingredients is of second-place importance to that to be gleaned from a complete knowledge of the origin &c of the water. The Medical Officer of Health is on the spot, he is trained to judge and estimate conditions affecting, or likely to affect the purity of a water supply, and he ought to be able and generally is quite fit to determine the value of the quantitative analysis of a water. I do not hesitate to declare that I look upon a chemical analysis as merely comfirmatory of a careful inspection, and from personal experience I have not found a water that could be justly condemned by chemical methods, but was also condemned by careful examination of the water course, or in case of a well, the surroundings, especially the flow of the ground water. The Medical Officer of Health is therefore the individual who is best fitted to report upon the suitability or unsuitability of any water supply. Uniform methods of analysis are desirable and the Society of Public Analysts and a Committee of the British Association on Chemical and Bacteriological Examination of Water and Sewage have issued Reports in favour of such an idea, which it is hoped will be followed by Analysts. In Germany, America, Australia &c the question of water supplies is not left as a local problem but is invested with the importance of being/

being a national question. In 1857 Karl Ehmann was commissioned by the Government of Wurtemberg to advise all Local Authorities on all questions of water supplies. A year or two later he was placed as the expert in a Department in charge of these matters.. Up to 1896 he had advised over 700 communities. In Bavaria schemes for 1488 places have been furnished by the Government up to 1898. The Grand Duchy of Baden in 1856 appointed three engineers for this purpose and in 1868 the work necessitated the appointment of three more. In rural districts in this Country the initial expenses connected with water supplies means in several places a considerable number of pence in the pound rental for 30 years, and entirely inadequate schemes are adopted, which require periodical tinkering and are never satisfactory, owing to the want in the initial stages of a competent experienced authority to guide and advise.

In Germany a code of directions for the purification of surface waters by sand filtration promulgated by the Minister for the Interior came into force on 13th.January,1899. Full details are given in the Jour.fur Gasbeleucht 1899 p.330. Rules are described regarding the water, its bacteriological examination, if filter defective what has to be done, and when filter out of use the conditions to be obtained before supplying for consumption. Rules for regulating flow, construction of filter &c &c are given/

given. How to take samples, the preparation of culture media &c &c are all given. These rules are to be obeyed not merely in the face of an epidemic, but at all seasons. Probably our own country will, in the course of time, see fit to follow such examples.

If the present state of chemical methods are not quite satisfactory in every respect, what can be said for the claims put forward by bacteriologists as to their science being the one to pronounce a true verdict on sewage pollution. A belief in the germ theory of disease, and the rapid advancement of bacteriological tech~~in~~ni gue naturally strengthens very much the hope that to bacterioscopic methods will the solution of the difficulties experienced in chemical methods ultimately tend. Disease is of vital origin and chemistry even in the third figure of decimals is unable to touch that point.

It was only after August 1881 when Koch explained in London his "plate" methods that bacterioscopic examinations of water were rendered possible. During the intervening 22 years much work has been done in this respect. The microbes in pure water, in stagnant or marshy water, in sea water and in sewage have been extensively studied and recorded by many observers. Much that has been recorded has been unverified, and much dogma has been stated that requires to be allowed to pass. The beginnings of every branch of knowledge of a practical nature generally exhibit such stages. Many of the most important researches into/

into conditions bearing directly on our subject have been undertaken by highly trained observers for the Local Government Board of England. I refer more particularly to Klein, Houston, S.Martin and Gordon's experiments. The want of a classification is a serious drawback, but the time is not yet ripe for more than a provisional one, and of these Professor Marshall Ward's, as detailed in the Journal of Pathology a year or two ago, is very helpful, and quite satisfactory till a more natural classification is devised. It is very confusing if not bewildering for an enthusiastic tyro, when he gets beyond the elementary stage, to read the contradictory opinions of distinguished workers. Here is a specimen which can be put in a syllogistic form. A de Simoni in the Centralbl. für Bakt 1900 p.426 shows that Bac. mucosus ozaena is the same organism as Friedlanders Pneumo-bacillus. Grimbert and Legros in Comptes.Rend.1900 p.1424 show that Bac aerogenes lactis is the same as Friedlanders Pneumo-bacillus. An Axiom of Euclid's allows even a non-bacteriologist to declare other two organisms to be identical. The Bac enteriditis sporogenes is declared by R.F.Hewlett (Trans.Path.Soc.1901 p.114) to be identical morphologically, culturally and in its pathogenic action with Achalme's Bac. of Acute Rheumatism, and such assertions of similiarity between what has been believed to be the casual organisms of different diseases is of the commonest occurence in bacteriological/

bacteriological literature. It must almost produce a bacteriological agnostic in one who has been taught dogmatically, but ultimately the feeling changes to one expressive of the little known and the much to know. The very prefix pseudo to so many micro-organisms is representative of our want of knowledge (pseudo-diphtheria, pseudo-tubercule bacilli, pseudo-tetanus bacilli, pseudo-pneumococci &c). Such terms are objectionable in many ways. A bacillus must either be the bacillus of Tuberculosis or not, and if it produces gross pathological signs and bacterioscopical appearances approaching those of the Tubercle bacilli, owing to that similiarity to name that which is probably an entirely different organism — pseudo-tubercle-is to hinder progress of knowledge. Prof. Farlow in his address as President of the Botanical Section of the A.A.A.S. at Boston in 1898 said specially referring to fungi "The best systematist is not he who attempts to make his species conform to what he believes to be the ideal of nature but he who availing himself of all the information which the histology, embryology and ecology of the day can furnish, defines his species within broad rather than narrow limits in clear and sharply-cut words which can be readily comprehended and do not force one to resort to original and perhaps single specimens to learn what the author of the species really meant". (American Naturalist,1898, p.675). Much of the divergence of results obtained by observers is probably/

probably due to want of uniformity in methods of procedure. That some uniform plan should be adopted is becoming more and more evident, so far at least as ordinary work is concerned. Special media will probably always be necessary as the outcome of experiment. At present it cannot be denied that much uncertainty exists regarding the correctness of the conclusions of even the most eminent experimenters in the domain of bacteriology, and only where it ~~assumes~~ practically assumes a question of national importance is any heed paid to their conclusions, outside the laboratories of similiar workers. Even clinical and pathological experience, as exemplified in Lord Lister and others at the Congress on Tuberculosis in London in July,1901,is sufficient for a time at least to overwhelm the laboratory researches of such an authority as Koch, the results obtained being often so contradictory. Such a state of affairs therefore engenders a confusion of mind in the reader that is most unsatisfactory. Even the titles given to papers by their authors are more often than not strangely misleading, and instead of being content to look upon their work as a contribution to the subject on which they write they boldly assert the discovery of something far beyond what can be legitimately deduced from the results recorded. For example in the Centralb. Bakt.1900 p.21 is an article by Dr Mankowski with the title "Procedure for Early and Rapidly distinguishing Cultures of Bac.Typhosus. from Bac.Coli." The medium for this/

this purpose is a decoction of mushrooms and some special stains which every subsequent observer has declared to be ineffective.

The powers of Bacteriology to discern the sewage contamination of water have been proclaimed to be of a high order. Dr Houston, in his Report on the "Chemical and Bacteriological Examination of the Washings of Soils" published in the 29th Annual Report of the Local Government Board 1899-00 p.523, states "Washings from soils enormously and dangerously polluted might up to a certain amount contaminate a water and not be chemically appreciable; yet the same contamination would yield to Bacterial Examination unequivocal proof of its objectionable origin." This statement is from a skilled Bacteriologist, but so much that has been written has been unverified that a sceptical attitude towards such statements may be pardonable. From the practical point of view of being able to trace the essential cause of a Typhoid Epidemic, Horrocks "Bacteriological Examination of Water" page 225 states that "it has been impossible, with one or two exceptions, to isolate the Bac. of Typhosus from water supplies during epidemics". These exceptions occured in epidemics in 1899 and 1901. At page 222, however, he ends a case where the Bac. Typhosus was isolated from water which had been drunk for three months although no enteric fever existed either in the Civil or Military population supplied by it. To invets such a discovery with an ~~act~~ air of importance/

importance, a theory has necessarily to be propounded that abortive forms are present in water, soil &c waiting to be endowed with the property of Pathogenic Action. The Bacterioscopic evidences of Sewage Pollution are Bac.Coli.,Bac.Enteriditis Sporogenes, and Streptococci. It has been shown that in Sewage a fair average sample shows 100,000 B.Coli. to each C.C. and from 10 to 1000 spores of B.Ent.Spor. in the same quantity. In ordinary domestic sewage the figures are very much higher. Klein and Houston in the 28th Annual Report of the Local Government Board, declare that one part of sewage in a thousand parts of water is scarcely detectable by delecate chemical tests, but by Bacterioscopic methods there would be no difficulty in detecting one part of sewage in 20,000 parts of water, by the B.Coli.tests, and so far as Bac.Enteriditis Sporogenes is concerned the delicacy of the test is enhanced so much that one part of sewage in 500,000 is detectable. It is not so much the actual presence of such organisms that is important. B.Coli.,it is said, is absent or nearly so from pure water. The term Coli.,however, is far from being a definite expression. It is applied to micro organisms that vary considerably, and the characteristics of the Escherich's Bac. Coli.Communis sometimes far from being realised in organisms grouped under the family name. So many varieties of Coli have been named that it is practically inpossible to follow descriptions. Stodart in the Journal of Pathology/

Pathology,1897, declares that in his experience, true Bac.Coli. are by no means so common as is generally supposed, and that they are at least as difficult to identify in pure culture as the Typhoid Organism. Houston confirms this opinion of Stodart by showing that B.Coli. appears to ~~be~~ somewhat readily die out even from polluted soils, and shows its almost total absence from virgin soils. From experiments carried out in Chichester, he was able in only two samples of soil taken from the neighbourhood of houses in which typhoid fever had recently occurred, to discover B.Coli. Chemical analysis showed the soils to be rich in organic matters. Although convinced of its powers of growth outside the body, he believes that under such conditions, in the course of a few weeks at most, it dies out. Its alleged ubiquity is therefore being assailed. It appears that many organisms included under the Coli group at present may be separate species, only our Bacteriological methods are not sufficiently advanced to discriminate between them. Mervyn Gordon declares of Coli that "instability is a characteristic family trait". Haslam (Jour.Path.1898) shows that "the pleomorphic forms of Coli range from small coccal-like forms to long thin rods and even spirals have been noted". He showed that in proteid solutions with ammon.tart. there were always many long forms; in glucose media short ones were in the majority. Food may thus be a

cause/

cause of pleomorphism, but as different sizes are always present in the same tube, it cannot be the only cause. Vincent Harris has also studied the great morphological differences of the Coli group, as found in the excretions of different animals. Escherich (the discoverer of the Colon Bacillus) recognised the remarkable pleomorphism it showed. Having seen it stated that Bac.Coli in pure water differ from those in polluted water in being devoid of Pathogenic Action, Dr J. Weissenfeld experimented to test this statement. He injected intra-peritonially into guinea-pigs some water from pure and some from infected sources. His experiments showed that Bac.Coli.could be obtained from almost every water if sufficient were taken, but failed to prove that a virulent Bac.Coli. was a test of its faecal origin. (Zeit-schrift fur Hygiene 1900 p.78) Louis Jenner Jour.Path. 1898) states that Coli from excreta are thicker and more opaque than Coli from pure water, but Larnelle got the transparent forms from the opaque by passage through the peritoneum, and other observers have obtained the same result by growth in milk. The effect of salt in media has been recorded by various observers and all seem fairly agreed on its producing alterations of shape and involution forms directly as the amount used. The reaction of the media affects the protoplasm of Bac.Coli as is shown by the fact that grown in a 3% alkaline medium

polar/

polar staining is obtained; but grown in an acid medium there is no polar staining (Escherich and Buchner).

In the Archive fur Hygiene 1899,p.192, we learn that prolonged cultivation causes the loss of flagella in at least micrococcus agilis and micrococcus citreus agilis although we further find that Zierler found, and Lehmann supported him, that Bac. Implexus had acquired motility in course of cultivation. Grumbert and Legros were able by cultivation to modify the function of Coli in two out of five varieties so far as the power to produce indol was concerned. Pakes (Brit.Med.Jour. 1901, p.958) states that Bac Coli varies in its fermenting action on Glucose and Glycerine. Some ferment glucose, some glycerine, some both, and some neither. Gordon (Jour.Path.& Bact.1897) has classified the varieties of Coli according to their actions and their flagella averages. They had previously been divided into motile and non-motile forms with sub-divisions sufficiently numerous to bewilder any one attempting to follow the classification. Gordon's classification was founded on experiments carried out by himself. In my own experiments Ihave found evidence to fully corroborate the value of the number and appearance of the flagella as a means of distinguishing between B.Coli and B.Typhosus, yet I cannot avoid concluding that Gordon's experiments do not show results that would warrant one in using them/

them as a basis for even temporary classification. His samples of coli were taken from normal stools, sewage, jaundic stool, mussel, water, milk, urine, and polluted water. Some of the coli produced gas, others did not, in ordinary gelatine shake and glucose gelatine shake. Some clot milk at 20 C. and 37 C. and others do not. Some clot at 20 C. and not at 37 C., and vice versa. In broth some produce indol, others do not. On potato the appearances ranged from a colourless to a brown growth. Some react acid, and some alkaline. The latter are rejected as not being coli, according to his classification. The flagella of his coli range from 1 to 10. Some of these organisms liquified gelatine, others did not, and the commencement of liquifaction ranged from the sixth to the thirty-sixth day. I do not think that many Bacteriologists would agree with Mr Gardomin regarding an organism which produced no clot in milk, no gas, and no indol as likely to be coli. None of the alkali produces clotted milk. This is said to be due to the production of an acid. What that acid is is as yet undetermined.

Apart from its value as an indication of the pollution of water, much of the interest that attaches to B.Coli arises from the difficulty Bacteriologists experience in readily distinguishing it from B.typhosus. Wathelet and others maintain, and the statement is universally accepted by compilers of works on Public Health/

Health, that B.typhosus is overpowered in the intestine, in sewage, in polluted soil &c by B.Coli. Pfuhl in Centralb.f.Bakt. 1899, p.49 records a series of experiments showing the commensalism of B.typhosus B.Coli, and Bacteria from garden soil grown on boiled potato. His experiments demonstrate that B.typhosus is not suppressed by other organisms competing on same medium. Remy in the "Ann.Instit.Pasteur" 1900, p.707,supports this latter view. He showed that B.typhosus and B.coli exhibited commensalism and reacted profoundly on each other, so that B.Typhosi were not agglutinated by Anti-typhoid serum as a result of the commensalism of the two organisms. Coli similiarly, he states, may be prevented from forming gas and indol. If an anti-serum is prepared from a coli growth it agglutinates the coli of the same descent as those from which it has been prepared, but it fails to agglutinate coli of another strain. Theoretically one would imagine that the specific nature of an organism should be found, if anywhere, in the vital results of its protoplasmic activity. A recent writer in the British Medical Journal hoped to show how this idea might be utilised in the separation of coli from typhosus. Taking advantage of the more rapid movements of the latter in broth, and using coli from which the anti-coli serum had been obtained, he was eminently successful in obtaining/

obtaining positive results, but with a loop of sewage inoculated with typhosus he found that his anti-coli serum had no agglutinative power over such coli, thus rendering his method absolutely useless.

Dr William Hunter (Lancet, 1901, p.613) describes a method of distinguishing a coli culture from a typhoid, by means of a solution of neutral red. Bac. coli reduces this substance, typhosus does not. Bac. enteriditis (Gart) reduces neutral red and Dr Hunter therefore believes it to be a coli. Wolff in the Centralb. f. Bakt. 1900, p.849, discusses the reducing powers of various micro-organisms. He shows that ~~anaer-~~ anaerobes possess the greatest reducing power, especially the organism of malignant oedema. He states that coli and typhosus are strongly reducing, anthrax and cholera faintly reducing, and tubercle not at all. ~~Methylen~~ they Methylen blue and litmus are substances that are also used as agents to show the reducing powers of bacteria They are easily reduced and again readily take up atmospheric oxygen. At present Hunter's method must be looked upon as one of the very many processes devised for separating Coli from Typhosus, and if this method can do it as Hunter says in 12-24 hours it will indeed be valuable.

Bac.Enteriditis Sporogenes. Since the discovery of this microbe by Klein in Oct,1895, it has been raised to a place of much importance as a test of experimental/

experimental pollution of water. In Klein's original description he says that it is not present in ordinary evacuations, and Andrews stated that it was not found in common diarrhoeal discharges. They regarded this organisn in the milk supply as the causa causans of the short sharp epidemis of diarrhoea which had occurred at St.Barts Hospital. Hewlett in the Jour. of the Mirroscopical Soc. 1899 declared this bacillus to be a normal inhabitant of the intestines, both in health and disease. Klein has since shown that sewage contains large numbers of its Spores - average 500 per c.c. (Houston 100 - 1000 per c.c.) and other observers have shown its practically ubiquitous character, thus verifying a conclusion deducted by Hewlett from the resistant nature of its Spores. Regarding the importance of this organism in water examination Houston states that its pressure "not only equals but far exceeds any known test chemical or bacteriological for inferring the wholesomeness or otherwise of a drinking water." But he also points out that the faculty possessed by it of sporing diminishes the value of its indicating recent pollution and therefore presumably dangerous pollution, but Horrocks that if the spores are found associated with "typical B.Coli the fact of sewage contamination may be considered sufficiently proved for all practical purposes". But B. Enteriditis Sporogenes has striking resemblances to other organisms and from the B.Butyricus of Botkin it

can/

can only be distinguished by innoculation experimants - an impossible kind of experiment for many workers. Houston believes that it does hot multiply in water. In a later report he speaks of this microbe as typical of excremental matters and declares it to be a certain delecate and easily applied test. At page 459 of the 29th Annual Report of the Local Government Board he however says "Contamination indicated by this microbe may have been recent, or it may have taken place months or even years previously." He accordingly proceeds "to search for another bacterial test which should affirm that organic matter of animal outcome had quite recently and only quite recently gained access to the water", He believes B.Coli to imply recent pollution, but Streptococci to imply extremely recent and therefore specially dangerous pollution.

Strepto-cocci. Strepto-cocci have long been recognised as being present in domestic sewage. Laws and Andrews showed that they were the commonest organism in the fresh sewage from St Barts Hospital. Houston was led to study the strepto-cocci and staphylo-cocci, but the latter he dismisses from further investigation "for the reason that they comprise hardy germs capable of persisting under conditions the reverse of favourable". Strepto cocci on the other hand only survive "when the conditions are almost ideally propitious, therefore their presence in soil, sewage or water shows recent association with an animal host", probably intestinal.. In crude sewage he estimates the average

number/

number of strepto cocci present to be 1000 per 1 c.c. Like coli they lose vitality and do not multiply to any great extent and consequently soon die out. He speaks thus of these micro organisms as a class, because in some special circumatances he has found that some strepto cocci are far from being delicate. Still his conclusion is that their presence in water is suggestive of recent pollution and therefore specially dangerous and when found "would go far to justify the bacteriologist in condemning asample of water as unfit for domestic use"; and directs that the search for these should therefore constitute an important part of the bacterioscopic analysis of potable waters.

Bacteriological Examination of Waters

Horrocks p.124 from experiments carried out by himself on this subject does not believe that strepto cocci indicate dangerous contamination. He found that the commonest organism which persisted in old sewage was strepto cocci and staphyococci. B.Coli had long before disappeared from the sewage and he reasons that likely so would have B.typhosus if present. The strepto cocci however rapidly disappeared when the old sewage was diluted with water. He therefore concludes that the presence in a water supply undoubtedly indicates a recent contamination, but the contamination is not necessarily dangerous unless the strepto cocci are accompanied by B.Coli. As these two opinions by competent observers are almost diametrically opposed further investigation is apparently/

apparently desirable, but in this instance so divergent views are expressed that the wonder is -- how can such divergencies happen. Dr Houston has either come to his very strongly worded conclusion without sufficient experiments or investigations, or Dr Horrocks may be simply making his statement after experimenting with certain old sewage and has assigned undue importance to the phenomena there observed, which are certainly not sufficient to justify such a sweeping generalization. Both cannot be right, although both may be wrong. All this tends towards developing a sceptical frame of mind, so far as the present claims of the science of bacteriology to show recent contamination of water by sewage. Houston's claim for bacteriology "that it is capable of detecting microbes characteristic of sewage under ordinary test conditions and can do so when sewage is from 10 to 100 times less than can be detected by chemical methods in ordinary use" is more than a justifiable deduction from his experiments. He carried out chemical analysis as well as bacteriological examination. Many of these with sufficient albumenoid ammonia to condemn them eight or ten times over (grossly polluted canal water &c.) were free from streptococci. A strongly worded claim like the above extract is altogether out of harmony with any subsequent admission that a negative result proves nothing. It is useless to give as an excuse the day of the week as being a probable cause of the want of success/

success, in connection with a water so polluted that any special sense (except hearing) would have pronounced it unfit for a water supply.

Typhosus. It would be impossible to conclude without referring to Bac.typhosus as an element in sewage contamination of water. Canstatt as long ago as 1847 stated that the stools of Enteric cases contained the poison. Budd also strongly supported this view. Long before bacilli were thought about investigators traced epidemics to water, milk &c. Murchison never departed from his belief in ~~ist~~ ulz origin from decomposing organic matter spontaneously, and many army surgeons at the present day are of the same belief. Various observers have shown that ingestion of putrid materials produce fever, diarrhoea and intestinal lesions. Eberth in 1880 discovered the bacillus which is generally believed to be the causal organism of the disease. Roux andRodet believe that B.Coli becomes changed into B.typhosus, although no one has been able to carry out the transition in vitrio. The law of Koch requiring the productions of the disease experimentally by culture is an impossible condition, as lower animals are not subject to typhoid. As yet there is only strong reason for accepting the Eberth-Gaffky bacillus as the cause of typhoid fever. Water is the generally accepted mode of conveyance of the organism and the two special conditions favourable to its distribution and growth are defective drainage and/

and contaminated water supplies. Some will listen to no other mode of conveyance than by water although Ballard, Gruest, Hart and others have collected information implicating food. Oysters through contaminated water have since Prof. Conn's celebrated Middletown epidemic had much notoriety as a cause of dissemination of the disease, and especially for sporadic cases as pointed out by Broadbent. ~~This~~ Flies in the Spanish-American and in our own South African Wars have been repeatedly certified as a means of dissemination of Typhoid Fever. Although for a time negatived by many observers it is just possible that Pettenkoffer may not have been so far away from hitting the right track for research when he stated that the poison of Enteric is not eliminated in a condition capable of communicating the disease directly, but that it must undergo changes in the soil, which changes are favoured by ground water. It is simply theory on the top of theory, but if the means of dissemination of the disease is as we generally believe it to be, some change must come over the Bac. typhosus in the soil, owing to some conditions as yet unknown which enables it to differ so much from laboratory experiments in glass tubes. The cause may not be ground water. The seasonal variation of typhoid shows it to be more prevalent in the late Autumn but so is scarlet fever, but ground water is not mentioned in connection with the latter disease. Buchanan thought he exposed the fallacy of Pettenkoffer's/

Pettenkoffer's ground water in suggesting that pollution of wells was a more probable origin. Baumgarten believes that in hot and dry seasons more dust is disseminated, which holds the bacillus of the disease, and so explains the greater prevalance. But other workers have shown that the chemical rays of sunlight speedily kill typhoid germs. Exposed in sterile water to sunlight they die in 1 hour. Germano showed that the bacillus quickly dies when completely dried in air currents. Even the discovery in the faeces of typhoid bacilli might not necessarily imply Enteric fever in the person. Discredit has attached to Diphtheria in this respect, by Bacteriologists certifying swabs as diphtheritic, which had come from normal throats, but exposed to diphtheria contagion. In experimental cholera infection of man no pathogenic action was produced although comma.bac. were found in the bowel evacuations. Metchnikoff found comma bacilli in the stools of healthy persons in absence of epidemic. Rumpel in Hamburg found comma bacilla in the stools of healthy persons during epidemic. Typhoid bacilli being found in water which were not producing any disease; an easy explanation might be that B.Coli have been mistaken for B.Typhosus. The number of individuals who have certified methods for separating Coli from typhosus is very great, and none if of any merit escapes criticism. Peppler last year carefully tested Piorkowski's method of growing and detecting typhoid bacilli, but concludes with the damning/

damning faint praise, that although of considerable merit it was not always satisfactory.(Centralb. Bakt. 1901, pp.879-81). Cambier in the Comptes Rendus 1901 p. 1442 describes a method for the isolation of B. typhosus owing to its greater motility. He was able to obtain the bac.typhosus from the Seine and the Marne and he claims it to be applicable to stools. In this paper he further reports the living together of coli and typhosi and in such circumstances the coli taken on the property of being agglutinated by typhoid anti-serum. Horrocks in the Brit.Med.Jour. 1900 p.1015 had previously made the statement that an organism that fails to agglutinate with typhoid-serum is not Bac.Typhosus but the converse is not true, all will admit, the importance of such a research if proved true would be very great, in connection with the discovery of typhoid bacilli in stools. Auerbach and Unger (Deutche Med. Wochenschr 1900 p.796 describe methods for isolating B. typhosus from blood and were successful 7 out of 10 times. By other observers the rose coloured spots had yielded the bacilli in a like proportion of cases. Others with apparently more bacteriological enthusiasm than common-sense or humanitarian feelings have thrust the needles of exploring trocars into the spleen in their zeal to demonstrate the bacillus.

Much is heard of the great variability of the colon bacillus, and a condition of mind is apt to be produced/

produced that B. typhosus is always more or less invariable. This is a part that offers ground for research of which I am to take the oppurtunity this summer. From the known experiments of Kraus, Hueppe, Karlinski, Bobrow &c the life of B.typhosus in water is remarkably short. Some as yet unknown variation is an essential condition in the production of disease epidemics by water. Meteorological conditions can only affect the temperature and the moisture of the soil, and in a modern watered and sewered town these cannot apply directly as an aetiological factor in the causation of an epidemic. Here an important link is awaiting solution.

Conclusion. I have not sought to introduce much detail in my paper, so as to enable me in the course of a thesis to touch broadly on the subject attacked. Many references to papers collected for this subject have been ommitted and much has been deleted otherwise.

It has not been my purpose to nicely balance opinion for and against, my purpose was rather to show that uncertainty exists so largely in reference to the contamination by sewage of a water that any opinion should be carefully guarded and no dogmatic statements are permissible. Although much assailed Chemical Analysis is of much importance in water contamination and although its processes may be capable of great improvement, it would be unwise to sit and wait till perfection is attained.

With Bacteriological researches one cannot but grant/

grant that during the last 20 years during which it has practically existed an enormous amount of research work has been carried out, and diversities of conclusions may be due to diverse methods and personal equations. Unless minute details are followed results are apt to disagree. In a class of say eight students, using the the same stains and following the same directions for staining flagella, only three or four will obtain decent specimens. Water from a mountain side flowing down a pebbly stream far from sources of any pollution the only examination necessary would be the physical characters and its hardness. From a stream flowing through a cultivated country all the tests or examinations possible should be taken advantage of, and after a personal examination of the sources and courses of the stream, the only person who should weigh up and pronounce the final judgement is neither the chemist or the bacteriologist but the Medical Officer of Health, from a collaboration of all the sources of information. The unwarrantable assumption of the past from single chemical analysis of a water, are still being continued in many cases owing to the analyst being demanded to furnish an opinion on the sample, and which opinion is taken by a Local Authority as a general conclusion on the quality of the water at all times. The Bacteriologist is not so often consulted except in the actual presence of an epidemic and the feat demended of him has been to discover the microbe/

microbe producing the epidemic. He has endeavoured by various methods to satisfy the tickled curiosity of the class of men who usually sit on Local Boards, and honestly admitting his want of success the wrath and derision of the peoples representative knows no bounds and is again rekindled when the Bacteriologist's account is presented. Several times this has happened in England, and any sensative worker had better leave such work alone, or for a time do what many bacilli do when finding themselves in uncongenial surroundings develop a skin like a spore. It should be clearly understood that an opinion on the Bacterioscopic examination of a water in other than highly trained hands cannot but be misleading and experience in this class of work is of more importance than even in chemical analysis. Single examinations may also be misleading. Developments are constantly occurring, and without being too sanguine of their correctness, the germ of further progress may be enlivened by even misstatements, and lead to research in the same or other directions which may themselves produce or originate fresh possibilities, or happily certainties. We can only hold firmly to that which is true, but to bacteriology I fix my faith, to be able to show in the future the recent contamination of water by sewage, but at present day I hold that a thorough examination of the actual water supply is an important factor in judging of this question and should be considered at least/

least of equal importance to a chemical or bacteriological examination.